

What is Claimed:

1. A cell comprising a nucleic acid molecule wherein said nucleic acid molecule comprises:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice region comprising a branch point, a pyrimidine tract and a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

2. A cell comprising a nucleic acid molecule wherein said nucleic acid molecule comprises:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;

- b) a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

3 A cell comprising a nucleic acid molecule wherein said nucleic acid molecule comprises:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 5' splice site;
- c) a spacer region that separates the 5' splice site from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

4. The cell of claim 1 wherein the nucleic acid molecule further comprises a 5' donor site.

5. The cell of Claim 1 wherein the nucleic acid molecule further comprises a safety nucleotide sequence comprising one or more complementary sequences that bind to one or more sides of the 3' splice region.

6. The cell of Claim 1 wherein the binding of the nucleic acid molecule to the target pre-mRNA is mediated by complementary, triple helix formation, or protein-nucleic acid interaction.

7. The cell of Claim 1 wherein the nucleotide sequences to be trans-spliced to the target pre mRNA encode a factor VIII polypeptide.

8. The cell of claim 1 wherein the nucleotide sequences to be trans-spliced to the target pre-mRNA encodes exons 23-26 of canine or human factor VIII protein or exons 16-26 of murine human factor VIII protein.

9. A cell comprising a recombinant vector wherein said vector expresses a nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice region comprising a branch point, a pyrimidine tract and a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and

- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

10. A cell comprising a recombinant vector wherein said vector expresses a nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

11. A cell comprising a recombinant vector wherein said vector expresses a nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA

expressed within the cell;

- b) a 5' splice site;
- c) a spacer region that separates the 5' splice site from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

12. The cell of claim 9 wherein the nucleic acid molecule further comprises a 5' donor site.

13. A method of producing a chimeric RNA molecule in a cell comprising:

contacting a target pre-mRNA expressed in the cell with a nucleic acid molecule recognized by nuclear splicing components wherein said nucleic acid molecule comprises:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice region comprising a branch point, a pyrimidine tract and a 3' splice acceptor site;

- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

under conditions in which a portion of the nucleic acid molecule is trans-spliced to a portion of the target pre-mRNA to form a chimeric RNA within the cell.

14. A method of producing a chimeric RNA molecule in a cell

comprising:

contacting a target pre-mRNA expressed in the cell with a nucleic acid molecule recognized by nuclear splicing components wherein said nucleic acid molecule comprises:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA; under conditions in which a portion of the nucleic acid molecule is trans-spliced to a portion of the target pre-mRNA to form a chimeric RNA within the cell.

15. A method of producing a chimeric RNA molecule in a cell

comprising:

contacting a target pre-mRNA expressed within the cell with a nucleic acid molecule recognized by nuclear splicing components wherein said nucleic acid molecule comprises:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 5' splice site;
- c) a spacer region that separates the 5' splice site from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

16. A method of claim 13 wherein the nucleic acid molecule further

comprises a 5' donor site.

17. The method of claim 13, wherein the chimeric RNA molecule

comprises sequences encoding a translatable protein.

18. A nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice region comprising a branch point, a pyrimidine tract and a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain;
- d) a safety sequence comprising one or more complementary sequences that bind to one or both sides of the 3' splice site; and
- e) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

19. A nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 3' splice acceptor site;

- c) a spacer region that separates the 3' splice region from the target binding domain;
- d) a safety sequence comprising one or more complementary sequences that bind to one or both sides of the 3' splice site;
and
- e) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

20. A nucleic acid molecule comprising :

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII pre-mRNA expressed within the cell;
- b) a 5' splice site;
- c) a spacer region that separates the 5' splice site from the target binding domain;
- d) a safety sequence comprising one or more complementary sequences that bind to one or both sides of the 5' splice site;
and
- e) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

21. The nucleic acid molecule of claim 18 wherein the nucleic acid molecule further comprises a 5' donor site.

22. The nucleic acid molecule of claim 18 wherein the binding of the nucleic acid molecule to the target pre-mRNA is mediated by complementary, triple helix formation, or protein-nucleic acid interaction.

23. The nucleic acid molecule of claim 18 wherein the nucleotide to be trans-spliced to the target pre-mRNA encodes a translatable factor VIII polypeptide and/or a marker protein.

24. The nucleic acid molecule of claim 18 wherein the nucleotide sequence to be trans-spliced to the target pre-mRNA encodes exons 23-26 of canine or human factor VIII protein or exons 16-26 of murine human factor VIII protein. 25. The nucleic acid molecule of claim 20 wherein the binding of the nucleic acid molecule to the target pre-mRNA is mediated by complementary, triple helix formation, or protein-nucleic acid interaction.

26. The nucleic acid molecule of claim 20 wherein the nucleotide

sequence to be trans-spliced to the target pre-mRNA encodes a factor VIII polypeptide and/or a marker gene sequence.

27. The nucleic acid molecule of claim 20 wherein the nucleotide sequence to be trans-spliced to the target pre-mRNA encodes exons 23-26 of the factor VIII protein.

28. A eukaryotic expression vector wherein said vector expresses a nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII protein pre-mRNA expressed within the cell;
- b) a 3' splice region comprising a branch point, a pyrimidine tract and a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA; wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

29. A eukaryotic expression vector wherein said vector expresses a nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to factor VIII protein pre-mRNA expressed within the cell;
- b) a 3' splice acceptor site;
- c) a spacer region that separates the 3' splice region from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

30. A eukaryotic expression vector wherein said vector expresses a nucleic acid molecule comprising:

- a) one or more target binding domains that target binding of the nucleic acid molecule to a factor VIII protein pre-mRNA expressed within the cell;
- b) a 5' splice site;
- c) a spacer region that separates the 5' splice site from the target binding domain; and
- d) a nucleotide sequence to be trans-spliced to the target pre-mRNA;

wherein said nucleic acid molecule is recognized by nuclear splicing components within the cell.

31. The vector of claim 28 wherein the nucleic acid molecule further comprises a 5' donor site.

32. The vector of claim 28 wherein said vector is a viral vector.

33. The vector of claim 32 wherein in said viral vector is an adeno-associated viral vector.

34. A composition comprising a physiologically acceptable carrier and a nucleic acid molecule according to any of claims 28-33.